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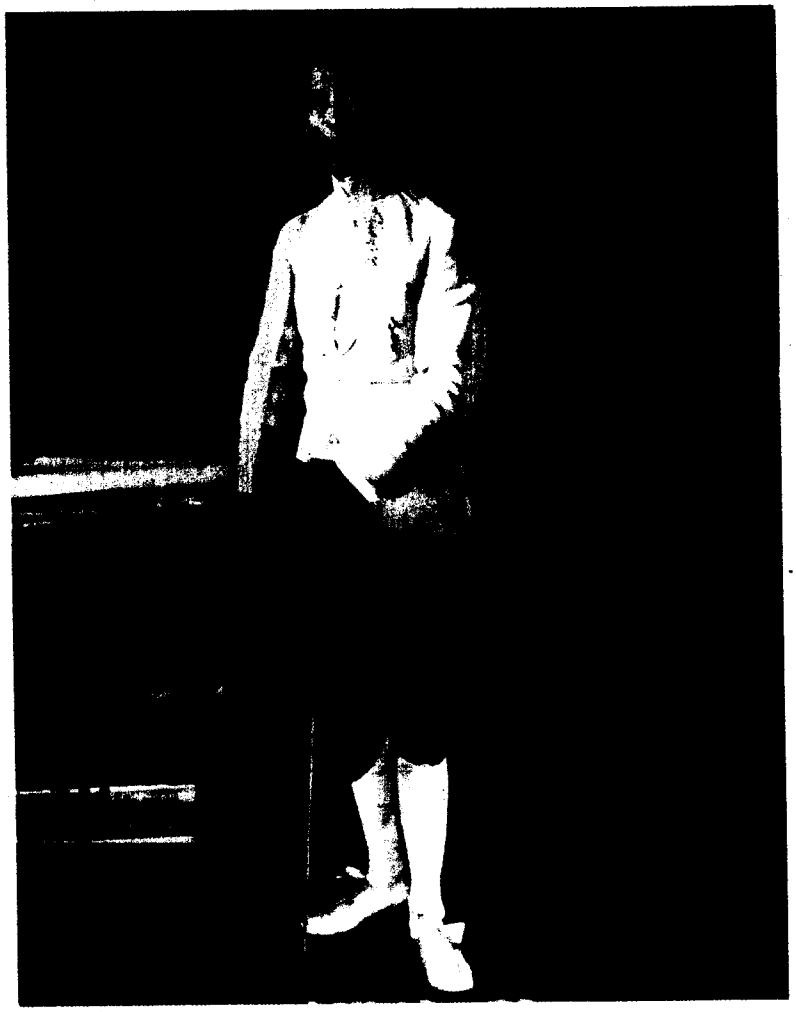

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Beyond the Human Genome: germs as genes*

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ABSTRACT Most research in infectious disease has been focussed on mechanisms of lethal virulence, and the corresponding host adaptations (mainly the immune system, invented 300 million years ago) corresponding thereto. Sparse attention has been given to the parasites' technology for sustaining themselves as chronic inhabitants, as domesticators of their macrobial hosts -- which includes their shared interest in muting virulence. Some startling example ssubstantiate the view that most encounters are driven by the parasites' entelic goal of joint survival. In some cases, the immune system of the host is manipulated to enhance resistance to superinfection by other competing invaders of the same or different species. Macrobial evolution, perhaps excepting pandemic disease of unusual ferocity (malaria) is just too slow to add more than a few nuances to the encounters. A year in microbial history matches all of primate, perhaps mammalian evolution. It may broaden our horizons if we think of the human as a superorganism, with an extended genome comprising: a) karyome -- chromosome set; b) chondriome -- mitochondria; c) microbiome -- entourage of microbial flora that share our body space.

KEYWORDS: genome, Karyome, chondriome, microbiome, microbes.

Medical science is imbued with a manichean view of the microbes' relationship to the human host. "We good; they evil." Microbes indeed have a knack for making us ill, killing us, and even recycling our remains to the geosphere. Nevertheless, in the long run, microbes have a shared interest in their hosts' survival: A dead host is a dead end for most invaders too. Domesticating the host is the better long-term strategy for pathogens. Were this not the case, large, complex multi-cellulars like ourselves could never have survived the evolutionary competition. The microbes have it all over us in pace of evolution, in sheer number, in promiscuity of exchange of genes.

The ascription of microbes to pathology has nevertheless pervaded the teaching of biomedical science for over a century, and left certain blind spots in our biological perspectives. Just for starters, the idea that bacteria had genes lagged the rest of genetics until mid-20th Century. That left an opening that initiated my own scientific career 55 years ago -- intertwined with the explosive developments in microbial genetics, and the implication of DNA as the genetic material, and eventually human genomics.

Next followed controversies over the role of extra-chromosomal particles in heredity. Some of these were plainly infectious viruses, and in the prevailing ideology that left them out of contention as genes, to the impoverishment of both genetics and virology. My introduction of the term "plasmid" just 50 years ago[1], helped many to climb out of that impasse. A given particle could be indeed be both gene and virus, at various stages of its life cycle. The genes of the plasmid could have either beneficial or deleterious effects on the host. Indeed, one of the first delineated plasmids, the F factor, is essential for the sexual cycle of *E. coli* bacteria. It has been plausibly argued that sex was invented by plasmids to facilitate their transmission to other host cells: that places sexually transmitted "disease" very early in the evolutionary time table. We may never know. Many mycoviruses have no other mode of transmission besides hyphal anastomosis, which can be regarded as a primordial sexuality.

We should take note of the life cycle of the retroviruses, which are unable to replicate until they have become integrated into host DNA,whereafter the RV replication is no other than the fairly standard transcription of host chromosomal DNA into RNA copies. The human genome is heavily populated with with "HERV", human endogenous retroviruses, and it appears that some of these have evolved further to take some part in our physiology, ironically most likely in the evolutionary refinement of the placenta.[2,3]

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* dedicated to the memory of Dr Vulimiri Ramalingaswami (1921-2001).

A recent commentary goes further:

"specific retroviral integrants ... niche functions that have conferred minor evolutionary benefits... For example, mice have adapted several ERV gene products [for] resistance to retroviral infection, and a retroviral insertion upstream [of] the pancreatic amylase gene may be responsible [for] humans, to taste starchy material as sweet"[4].

We have every reason to expect that, our most notorious retrovirus, HIV, will find a way to lodge itself in the germ line as well -- we have no idea what pathway the other HERV's used to reach that target nor the longterm consequences of this further evolution.[5]

Together with its symbionts/parasites, we should think of each host as a superorganism with the respective genomes yoked into achimera of sorts. To help dramatize this view of the human as a superorganism, human genome PLUS scores or hundreds of ancillaries, I propose the term "microbiome".

The microbiome would entail the entourage of microbial flora with which we share our body space, perhaps intracellularly as endosymbionts, but also on our skin, gut lumen, mucosal surfaces, and elsewhere. Each of these components can have an important impact on the outcome of our encounters with infection (and reinfection), as well as on nutrition and other phenotypes. The term "microbiome" can be read in two ways. One is "microbiome" referring to the small biotic community that defines each of us as individuals. "Microbiome" would be the collective set of microbial genomes which add still more spicy condiment to the goulash of our total DNA. There is even historic traffic between them, as we discover DNA sequences in our karyome that must be insertions from microbial sources. The chondriome, and its cousin in green plants, the (chloro)plastidome, are remnants of ancient invasions of the proto-eukaryotic cell by bacteria.[6]

The mitochondria, can then be regarded as the most successful of all microbes. They reside inside every eukaryote cell (from yeast to protozoa to multicellular organisms), in which they provide the machinery of oxidative metabolism. These examples reveal how far collaboration between hosts and infecting microbes can go, in the mutual interpenetration of their genomes.

Well evolved endosymbioses are rife among invertebrates, providing headlamps for deep sea squid, sources of nutrients for innumerable insects, and modulating sex ratios in fruitflies.[7] Transovarial lineages of rickettsia provide reservoirs for some zoonotic infections of humans, in the example of tsutsugamushi fever.[8]

The human as a superorganism, has an extended genome, comprising as far as we know:

a) karyome -- chromosome set
b) chondriome -- mitochondria
c) microbiome -- entourage of associated microbial flora

The components of the microbiome are promiscuously exchanging genes with one another, and occasionally with the karyome. The superorganism, the symbiome, becomes a unit of natural selection at one level, reminding us how tribes and other cohorts, have also been regarded as superorganic at more complex levels of social organization. We are also reminded of the arbitrariness of the boundary demarcations by which we choose to define an "organism".[9] The entire biosphere comprises more or less tightly coupled genomes exchanging nutrients and transferring solar energyin intricate webs: we marvel how a single primordial cell, beyond the awesome fact of its origination, could have survived the interval to the organization of that ecoweb.

The human microbiome is a poorly catalogued ensemble, of which the majority of entries have yet to be cultivated and characterized; not to mention its dynamics in the course of development, disease, response to nutritional and pharmaceutical intake. One of the most useful byproducts of the Human Genome Project has been the successful sequencing of scores of microbes, a list which is being extended weekly. The comparative genomics of microbes has already been of untold value in pinning down their evolutionary relationships, and then in providing hundreds of provocative leads for epigenetic insight and therapeutic targetting.[10]

To digress briefly to germs in pathogenetic processes: In the short run, the infected host is in fact at metastable equilibrium: the balance could tip toward favorable or catastrophic outcomes. Most successful parasites travel a middle path. It helps for them to have aggressive means of entering the body surfaces and radiating some local toxicity to counter the hosts' defenses, but once established they also do themselves (and their hosts) well by moderating their virulence.

Better understanding of this balancing act awaits further research, with a moderated biological perspective. And that may take a shift in priorities. For one, research has focused on hypervirulence. Studies into the physiology of homeostatic balance in the infected host qua superorganism have lagged.

Yet the latter studies may be even more revealing, as the burden of mutualistic adaptation falls largely on the shoulders of the parasite, not the host. This lopsided responsibility follows from the vastly different evolutionary paces of the two. But then we have our wits, it is to be hoped, for drafting the last word.

To that end, we also need more sophisticated experimental models of infection, which today are largely based on contrived zoonoses (the migration of a parasite from its traditional host into another species). The test organism is usually a mouse, and the procedure is intended to mimic the human disease process. Instead, it is often a caricature.

Injected with a few bugs, the mouse goes belly up the next day. This is superb for *in vivo* testing of an antibiotic, but it bears little relation to the dynamics of everyday human disease.

I suggest that a successful parasite (one that will be able to remain infectious for a long time) tends to display just those epitopes (antigen fragments that stimulate the immune system) as will provoke host responses that a) moderate but do not extinguish the primary infection, and b) inhibit other infections by competing strains of the same species or of other species. According to this speculative framework, the symptoms of influenza evolved as they have in part to ward off other viral infections, and in a complex way to mitigate its lethality and promote its dissemination to other hosts. The extended genome of a given human will then fluctuate with episodes of infection and disease, the traffic of different contenders for our body space. Viral agents which confer life-long immunity to survivors have evidently practised a kind of gene therapy, by one hypothesis, planting genomic residues in some long-lived body cells. If that is so, we have evidently been indulging in genetic engineering since Jenner's experiments with vaccinia, art emulating nature.

May we not supercede the past millenia's metaphor of war for describing the relationship between people and infectious agents. A more ecologically informed metaphor, which includes the germs'-eye view of infection, might be more fruitful.

This is not to pretend that our relationship with the microbiome is devoid of proximal conflict and turbulence, no more than do the human cells of a given body always follow rule of benevolent social order. Besides explosive epidemic outbreaks, we have to contend with the likelihood that many common chronic diseases may have microbial etiologies. This is well substantiated for some forms of arthritis and gastric ulcer, and under grave suspicion for atherosclerosis and some neurological disorders. For any prevalent, imputedly constitutional, disease one wonders what are the evolutionary drives to sustain susceptibility genes in the population -- say for depression? In these cases, one might argue that the genetic drive operates through an aggressive parasite, overtaking the evolutionary pace of the host.[11]

Another relevant issue that can be recast in an ecological mold is the rise in popularity of antibacterial products. This is driven by the popular idea that a superhygienic environment is better than one with germs -- the "enemy" in the war metaphor. But too much antibacterial zeal could wipe out the very immunogenic stimulation that has enabled us to cohabit with microbes in the first place. We can anticipate many phenotypes of microbiomal modulation of infection. To begin with, this is where we look most closely, and it makes sense in terms of inter-microbial competition. The adverse side-effects of antibiotic therapy - most notoriously Clostridium difficile take over after clindamycin - are an immediate pointer. The many immunological cross-reactions, especially among carbohydrate capsular antigens, cannot but have some role in major outbreaks. Milkmaids' fair complexions were (apocryphally) credited with inspiring Jenner to the discovery of vaccination against smallpox. We have some hint of viral interference from recent reports about hepatitis GBV-C being negatively correlated with HIV progression.[12]

In addition there are scattered findings of other nutritional and developmental roles of microbiomal diversification. Rather than my attempting a critical review in the brief time available, it will suffice to scan the appended bibliography, the papers having self-explanatory titles.

I do want to acknowledge my debt to Rene Dubos[13] for his inspiration in this line of thinking, which has other roots in the work and thought of Metchnikoff, Theobald Smith, McFarlane Burnet and many others

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